

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

**(19) World Intellectual Property Organization
International Bureau**



(43) International Publication Date
30 June 2005 (30.06.2005)

PCT

(10) International Publication Number
WO 2005/059157 A2

(51) International Patent Classification⁷:

C120

(72) Inventors: and

(21) International Application Number:

PCT/US2004/041569 ✓

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(22) International Filing Date:

10 December 2004 (10.12.2004) ✓

English

(26) Publication Language:

English

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(30) Priority Data:

Priority Data: 60/529,407 11 December 2003 (11.12.2003) ✓ US
60/532,506 26 December 2003 (26.12.2003) ✓ US

(81) **Designated States** (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG,

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[Continued on next page]

(54) Title: METHODS AND COMPOSITIONS FOR USE IN PREPARING HAIRPIN RNAs

Step 1)

Step 2)

Step 3)

Step 4)

Step 5)

Step 6)

Final vector

Transcribed

(57) Abstract: Methods and compositions for producing hRNA, e.g., shRNA, expression modules for specific target nucleic acids are provided. In the subject methods, an initial nucleic acid, e.g., dsDNA, synthetic DNA, etc., corresponding to the target nucleic acid of interest is converted to an intermediate nucleic acid. The resultant intermediate nucleic acid, following an optional size modification step, is then converted to a linear dsDNA that includes at least one copy of the hRNA expression module of interest, or a precursor (i.e., pro-shRNA expression module) thereof, where in certain embodiments conversion may include amplification. Also provided are reagents, systems and kits for use in practicing the subject methods. The subject methods and compositions find use in a variety of different applications.



PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

— without international search report and to be republished upon receipt of that report

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(84) **Designated States (unless otherwise indicated, for every kind of regional protection available):** ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, MC, NL, PL, PT, RO,